

EMBERIZID DIGESTIVE TRACT
LENGTH AND WEIGHT DYNAMICS

by

MICHAEL EDWARD BARNES

B.S., South Dakota State University, 1985

A THESIS

submitted in partial fulfillment of the
requirements for the degree

MASTER OF SCIENCE

Division of Biology
KANSAS STATE UNIVERSITY
Manhattan, Kansas

1987

Approved by:

A handwritten signature in dark ink, appearing to read "R. D. Real", is written over the printed name.

Major Professor

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ACKNOWLEDGMENTS

Sincere thanks are extended to Greg Farley for showing me the proper way to capture birds, and to Fred Wilson for teaching me how to maintain the birds in captivity. The invaluable moral support provided by Greg, Fred, and Sam Kruckenberg will always be remembered. The humor and statistical assistance provided by Ken Kemp are also much appreciated.

By letting me walk the railroad tracks during the Christmas bird count, John Zimmerman deserves credit for helping me locate my best junco netting location. His support during my stay in Kansas has been more than gracious. The advice of John Briggs and Elmer Finck was both useful and entertaining. Also, Greg Wells and the secretarial staff deserve acknowledgment for their indirect contributions to this work.

I thank Dr. R. J. Robel for the financial support of my research endeavors, including the use of a vehicle, environmental chambers, mist nets, and a computer. His taking time to edit this thesis is also acknowledged.

Lastly, I thank my wife, Nancy, for all of her help. Although assisting with a number of tasks, including mist netting, cage cleaning and evisceration, her most important contribution in this work was helping

me to keep my sanity throughout it all.

For my parents, Ola and Donna Barnes,
who taught me the value of an education,
and for Mary.

INTRODUCTION

With the start of Leopold's (1953) discovery of variation in California quail (Callipepla californica) intestinal morphology, research has concentrated on how the digestive tract changes in response to diet in birds. Most of this work has been with gallinaceous birds and waterfowl, because of the economic importance of these two groups. One of the major objectives of this study was to attempt to document experimentally the ability of the emberizid gut to morphologically modify itself, depending on the diet that the organism faces.

Other important, previously unanswered questions, concern how the sampling and evisceration techniques employed might influence measurements of gut morphology. For example, although it is well established that avian body weights fluctuate throughout the day (Bhatt and Chandola 1982, Kontagiannis 1967), no attention has been given to daily fluctuations in the length and weight of the digestive tract. Also, when specimens are collected, evisceration occurs at a wide range of times after death, with all of the data pooled for later analysis. No determination of the effect of such time lapses on gut morphology has yet been reported. To try and ensure that the morphological changes reported in the literature were caused by diet, and not by some data collection procedures, the determination of the effects

of several sampling parameters was another primary objective of this study.

LITERATURE REVIEW

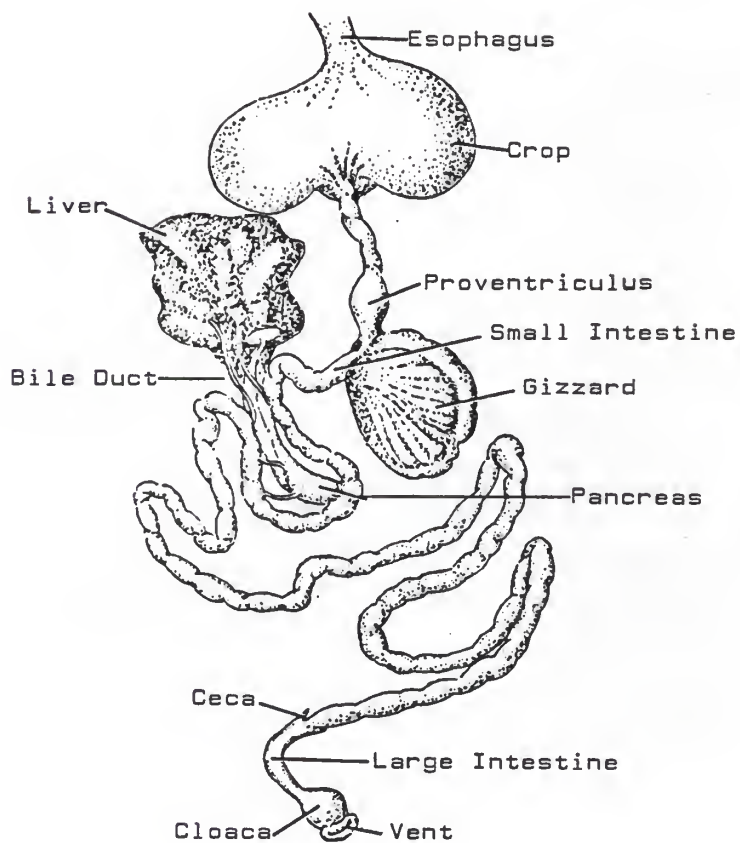
The avian digestive system as a whole is extremely plastic. The degree of development of certain structures has been reported to vary both inter- and intraspecifically, with differences in development attributed to food habits (Wallace and Mahan 1975). In this review, the general form and function of the avian digestive tract will be addressed first. A review of the morphological changes of the gizzard, liver, intestine and ceca that have been attributed to diet will then follow.

Gross Morphology

The structures of the avian digestive tract and the associated digestive organs are shown in Figure 1. Food enters the buccal cavity and is passed intact into the esophagus. Mucous epithelium in the mucosa lubricates the food (Pettingill 1970) and the muscular layers propel it down to the glandular stomach, the proventriculus. In some species, the esophagus may temporarily expand for food storage, or it may have a permanent expansion or diverticulum called a crop (Ziswiler and Farner 1972). The proventriculus provides for initial food breakdown with its gastric secretions (Wallace and Mahan 1975) and may have other functions (Ziswiler and Farner 1972).

Two thick, opposed, lateral muscles and two thin

Figure 1. General Avian Digestive System



From Wallace and Mahan 1975.

anterior and posterior intermediary muscles, are the major components of the muscular stomach, the gizzard (Bradley 1950, Hill 1971, Klem et al. 1983, 1984). The gizzard receives food from the proventriculus. A hard koilin lining, not keration as suggested by Welty (1982) but a polysacccharide-protein complex (Englitis and Knouff 1962), is formed inside the gizzard by mucous epithelial cell secretions. The koilin lining functions as a protective coating and an abrasion surface during food maceration and acid protelytic digestion (Ziswiler and Farner 1972). Small stones (grit) may be present in the gizzard to aid in mechanical breakdown, or as a source of certain minerals.

Gizzard contents are passed into the small intestine, the primary site of food digestion and absorption. The small intestine receives bile from the liver which may, depending on the species, be stored in the gall bladder (Wallace and Mahan 1975). The liver has other nondigestive functions, such as lipid and glycogen storage (Ziswiler and Farner 1972) and is derived embryologically from the digestive tube (Van Tyne and Berger 1976). Pancreatic enzymes are discharged into the small intestine as well.

No definable histological demarcations occur along the length of the small intestine of birds, unlike those of other vertebrates (Gier and Grounds 1944, Hodges

1974, Ziswiler and Farner 1972). The small intestine mucosa is well developed for absorptive and digestive purposes (Klem et al. 1983), and the intestinal epithelium is generally replaced every 48 hours (Hill 1971).

A pair of blind end projections, the ceca, arise at the junction of the small and large intestines. The ceca function as fermentation chambers (Gasaway 1976a, 1976b, McBee and West 1969), and also reabsorb water (Gasaway et al. 1976) and digested proteins (Welty 1982).

The large intestine is relatively straight and short, and may function in water reabsorption (Ziswiler and Farner 1972). Histologically the large intestine varies from the small intestine only by the presence of large numbers of mucous secreting goblet cells (Hodges 1974, Klem et al. 1983, 1984). The cloaca is the terminal portion of the digestive tract, and functions for the digestive system as a waste storage center (Welty 1982), in addition to its other non-digestive functions.

Variation in Digestive Organ Morphology

Gizzard

Interspecific variations in the character of the gizzarde are considered by Ziswiler and Farner (1972) to be dependent on the texture of the diet. Wallace and

Mahan (1975) use this reasoning in generalizing that insectivorous birds have smaller gizzards than non-insectivorous birds, despite the influence of body size. However, Thomas (1984) and Moss (1983) found a correlation between gizzard weight and body weight in northern galliformes.

The variation in gizzard size among five duck species of the same genus (Bucephala spp.) was hypothesized to be inversely related to the amount of fiber in the diet (Kehoe and Ankney 1985). Lazareff (1949) maintained that no relationship between stomach structure and food consistency was evident histologically.

It has not been established if the change in gizzard size between species is due to dietary differences, or is simply due to the interspecific differences in body size.

Intraspecific variations in gizzard morphology appear to follow similar patterns as interspecific differences, generally being reported to change with changes in feeding or body weight. Seasonal variations in intraspecific gizzard size were illustrated by Spitzer (1972), who found that with a dietary shift from insects to seeds, gizzards became more muscular, developed a koilin lining, and contained grit. Hanssen (1979a) and Pendergast and Boag (1973) found similar

seasonal fluctuations in northern gallinaceous bird gizzard weights. Seasonal differences could also be a function of photoperiod or other uncontrolled variables however.

Miller (1975) demonstrated experimentally a positive correlation between gizzard weight and dietary fiber in mallards (Anas platyrhynchos), a correlation that was supported in the field by Whyte and Bolen (1985). Japanese quail (Coturnix coturnix) changed gizzard size in less than one week in response to changes in dietary fiber (Savory and Gentle 1976b). Gizzard weight decreases in the winter were attributed to dietary shifts from aquatic vegetation to low fiber corn in gadwalls (Anas americana) (Paulus 1982) and wood ducks (Aix sponsa) (Drobney 1984). Herd and Dawson (1984) and Hanssen (1979a) found wild birds had larger and heavier gizzards than captive birds of the same species.

Canada geese (Branta canadensis) (Raveling 1979) and sage grouse (Centrocercus urophasianus) (Hupp and Brown 1984) showed significant declines in gizzard weights during reproduction, presumably due to hypophagia. Snow geese (Chen caerulescens) gizzard weights fluctuate over winter, mostly due to body weight fluctuations (Hobaugh 1985). Conversely, Egyptian geese lose 19% of their body weight during molting, but the

gizzards of molting birds are 60% heavier than those of non-molters (Halse 1984). Kirkpatrick (1944) could find no changes in gizzard weights over time in adult ring-necked pheasants (Phasianus colchicus).

The effect of the time after death and the time of day the birds were sampled was not addressed in any of the afore mentioned studies and could have been responsible for the observed intraspecific changes.

Grit is often found in the gizzard, and the necessity of grit in the avian diet has been an area of controversy (Wallace and Mahan 1975). Herd (1985) contends that grit is deliberately ingested to aid in the mechanical breakdown of the food. Such deliberate ingestion has been observed in Harris' sparrows (Zonotrichia querula) (Graul 1967). Wallace and Mahan (1975) suggest that some grit is taken accidentally during feeding, which could explain the high incidence of sand in shorebird gizzards (Reeder 1954). Accidental ingestion would not explain the presence of grit in the gizzards of many caprimulgids however (Jackson and Mengel 1970).

Both young and adult bobwhites (Colinus virginianus) on a gritless diet survived as well as did those with grit (Nestler 1946). However, when domestic chickens (Gallus gallus) on a seed diet are given grit, a 10% increase in metabolic efficiency occurs (Welty 1982).

Beer and Tidyman (1942) contend that in the absence of grit, gallinaceous birds can use the seeds as abrasives against each other.

Water soluble minerals in the grit could provide potential benefits to the bird (Robel and Bisset 1979). Sadler (1961) documented female pheasant preferences for calcium bearing, limestone grit over other noncalcareous grits, during egg laying. A similar situation was found in Anna's hummingbirds (Calypte anna) (Verbeek 1971).

The effect of grit on gizzard morphology has not been directly addressed. Spitzer (1972) indirectly observed possible grit influences, but different feed, daylength, and time sampled compounded his results.

Liver

No seasonal fluctuations in liver weights could be found in ring-necked pheasants (Kirkpatrick 1944), ruffed grouse (Bonasa umbellus) (Thomas et al. 1975), or spruce grouse (Dendragapus canadensis) (Pendergast and Boag 1971). Whyte and Bolen (1985) also noticed no differences in the winter liver weights of mallards. Oakeson (1953, 1956) however, reported seasonal variation in avian liver weights. In both migratory and nonmigratory races of the white-crowned sparrow (Zonotrichia leucophrys), she discovered an increase in liver weights in the winter, with the lowest weights occurring in the late spring after the birds had already

established territories and mated.

Early spring increases in liver weight were attributed to hyperphagia in prebreeding, female rock ptarmigan (Lagopus mutus) (Thomas and Popko 1981), male sage grouse (Hupp and Brown 1984), female wood ducks (Drobney 1984), female willow ptarmigan (Lagopus lagopus) (Pulliainen and Tunkkari 1984), and snow geese (Ankney 1977). Decreases in liver weight occurred in snow geese (Ankney 1977) and wood ducks (Drobney 1984) during incubation. Canada geese liver weights decrease prior to and during reproduction (Raveling 1979).

Drobney (1984) and Pulliainen and Tunkkari (1984) noted a negative relationship between dietary fiber content and seasonal liver weight fluctuations. Liver glycogen levels change in the early embryonic and growth phases of the domestic chicken (Freeman 1969), and during adulthood in willow ptarmigan (Pulliainen and Tunkkari 1984). Glycogen fluctuations could not explain adult willow grouse liver weight fluctuations however (Pulliainen and Tunkkari 1984). Fat increases did not totally account for seasonal liver weight changes either (Oakeson 1953, 1956) and water and protein content remained unchanged (Pulliainen and Tunkkari 1984). Liver tissue must therefore either increase or hypertrophy (Pulliainen and Tunkkari 1984).

Seasonal liver weight fluctuations have not been

studied in a controlled environment, and could be due to any number of other variables than diet.

Daily body lipid variations were observed by Odum and Perkinson (1951), leading Fisher and Bartlett (1957) to examine variations in liver size after a night of fasting. Red-winged blackbirds (Agelaius phoeniceus) and European starlings (Sturnus vulgaris) exhibited declines of greater than 30% in liver weight overnight (Fisher and Bartlett 1957). Male liver weight decreased faster than female liver weight in fasting starlings, but the relative proportion of liver as a percent of body weight remained the same (Stegeman 1954). Liver weight also increases with increasing body weight in white-crowned sparrows (Oakeson 1954, 1956).

Glycogen levels fluctuate daily (Dolnick and Blyumental 1967) and remain depressed in the absence of food (Fisher and Bartlett 1957). Glycogen levels could be influenced by collection time though (Pulliainen 1985). Liver fat peaks at the end of the daily light period in premigratory chaffinches (Fringilla coelebs) (Dolnick and Blyumental 1967) and cycles even during fasting in red-winged blackbirds (Fisher and Bartlett 1957). Body fat and liver weight are inversely related in migrant chaffinches, and liver weight increases during the day throughout the year (Dolnick and Blyumental 1967).

Liver weight fluctuations have not have never been followed diurnally.

Small Intestine

Flesh eating birds are generally considered to have shorter and smaller intestines than seed eaters (Pettingill 1970, Pulliainen et al. 1981, Wallace and Mahan 1975) and fish eaters (Stone et al. 1978, Welty 1982), although Ziswiler and Farner (1972) contend that the surface area in the intestinal lumen does not change. Differences attributed to diet, such as the 15 m long small intestine of an ostrich compared to the 5 cm long intestine of a hummingbird (Wallace and Mahan 1975), are likely due mainly to body size differences, as Thomas (1984) found with three gallinaceous species.

Leopold (1953) and Kehoe and Ankney (1985) suggest interspecific intestinal length differences are due to differences in the amount of fiber in the diet. Increasing dietary fiber appears to be correlated with increasing small intestine length, but such a correlation could be influenced by sampling procedures.

Small intestine lengths vary as much within a species as they do between species (Moss 1983). Despite such variation, the literature abounds with examples of suspected dietary influences on small intestine length.

Using rats, Brownlee and Moss (1959) experimentally illustrated that small intestine length increased with

increased fiber in the diet. Davis (1961) was the first to show that in the same species of birds, rufous-sided towhees (Pipilo erythrophthalmus), small intestine length increased during the winter. This increase was attributed to a change from eating low fiber insects to high fiber seeds, but could also have been due to daylength, time sampled, or temperature. Lewin (1963) discovered winter lengthening in the small intestine of California quail.

After Miller (1975) found that mallard small intestine length and weight were related to dietary fiber and the amount of food consumed, several field investigations followed. Snow goose (Ankney 1977), Canada goose (Raveling 1979), and wood duck (Drobney 1984) small intestine lengths were found to vary throughout the year in these field studies. The intestinal length changes were again attributed to the amount of food consumed and dietary fiber, but several other factors could have influenced small intestine length. The action by which food consumption or fiber could have influenced small intestine length is not known, but McLandress and Raveling (1981) suggest that Canada geese in the spring consume the extra protein and water required for hypertrophy of the small intestine.

Gallinaceous bird small intestines have also been much studied. Moss (1972) noticed that captive red

grouse (Lagopus lagopus) eating mostly mash had significantly shorter intestines than wild birds eating mostly fibrous heather. He suggested that diet was the primary determinant of small intestine length, although several other factors could have determined the differences he observed. Moss (1977) also suggested that the increase in small intestine length in wild grouse over captive birds was responsible for the wild birds ability to digest heather more efficiently. Hanssen (1979a) reported that the small intestine micromorphology, length of villi and tissue layer widths, remains the same between captive and wild grouse, although captive grouse contain higher numbers of bacteria in their small intestine than wild grouse (Hanssen 1976b).

Several grouse and ptarmigan that have been examined change from a low fiber summer diet to a high fiber winter diet (Moss 1974, Pendergast and Boag 1970) that has been suggested to cause a corresponding increase in small intestine length (Moss 1974, Pendergast and Boag 1971, 1973, Thomas 1984, Pulliainen and Tunkarri 1983). This cause and effect relationship between the type of food consumed and small intestine length does not hold true for all galliformes, as Hupp and Brown (1984) and Kirkpatrick (1944) could find no change in adult male sage grouse or ring-necked pheasant

intestinal morphology in the spring. The discrepancies in the literature may be explained by different sampling methods and times.

Savory and Gentle (1976b) produced changes in the small intestinal lengths of Japanese quail in less than one week when additional fiber was added to the diet, but could see no difference in the rate of passage times in different length intestines (Savory and Gentle 1976a). The histology of the small intestine was also unaffected by diet (Gentle and Savory 1975) indicating that small intestine length increases were due to stretching of the small intestine. The time required for such stretching to occur is unknown.

Hormones have been shown to dramatically change intestinal length by inducing hyperphagia, such as in adrenalectomized rats (Haeger et al. 1953, Levin 1965) and rats where diabetes was artificially induced (Levin 1969). Though unnatural, these studies indicate that food consumption directly affects small intestine size. Levin (1969) suggests that the amount of luminal nutrition causes an increase in small intestine size, but this is unlikely given the lack of histological differences from birds on different diets (Gentle and Savory 1975, Hanssen 1979a).

The amount of food consumed is a factor that changes gut size (Fell 1969) and may be more important

than crude fiber in determining small intestine morphology according to Pulliainen (1976) and Pulliainen and Tunkkari (1983). Food consumed and dietary fiber are naturally related though, and the effects of either of these factors has never been separated from the other. Timing and duration of feeding changes seasonally (West and Meng 1966), and also likely influences small intestine length as well, although this has never been analyzed.

Sex and age differences in gut morphology are species dependent (Gier and Grounds 1944, Leopold 1953, Pendergast and Boag 1973), and geographic variation occurs as well (Amanova 1977, 1978). How these results are affected by sampling time, both daily and seasonally, has not been addressed. Nisbet et al. (1963) noted daily fluctuations in the digestive tract contents, but did not comment on the significance of such fluctuations on small intestine length.

Ceca

The presence of ceca at the junction of the large and small intestine is not a universal feature of the avian class (Pettingill 1970, Welty 1982). In those families where ceca are present however, diet may affect their morphology.

A causal relationship between dietary fiber and cecal size was first proposed by Leopold (1953). When

placed in captivity, red grouse ceca decreased in length by 50%, a decrease attributed to the reduction of heather in the diet during captivity (Moss 1972). Captive Japanese quail fed large amounts of fiber showed significant length and weight increases in one week (Savory and Gentle 1976b). Miller (1975) noted that in captive mallards, the ceca were most sensitive digestive organ to dietary change, increasing in length with increasing fiber in the diet. These studies indicate that fiber some how affects cecal length, but do not speculate as to the mechanism of the action.

The literature is replete with examples of naturally occurring changes in cecal morphology. Rock ptarmigan (Gasaway 1976a, Moss 1974), willow ptarmigan (Pulliainen and Tunkkari 1983, Thomas 1984), spruce grouse (Pendergast and Boag 1973), wood ducks (Drobney 1984), and gadwalls (Paulus 1982) have all been shown as having seasonal variations in cecal size. Whether such changes are due to changing diets or some other seasonal factor remains to be determined. Sage grouse cecal size does not change however (Hupp and Brown 1984).

Cecal changes in passeriform birds has not been examined; daily fluctuations in cecal size has not been addressed either.

Large Intestine

Despite the fact that Ziswiler and Farner (1972)

contend that large intestine length reflects diet, few differences in large intestine length have been detected with changing diets. Miller's (1975) data shows that the large intestine is the least sensitive of all the digestive organs to dietary changes, although it did increase in length with an extreme increase in dietary fiber. Leopold (1953) could find no difference in large intestine length between two races of California quail that exhibited other digestive organ differences. The large intestine of male rufous-sided towhees increased in length during the winter, while the large intestines of females, supposedly consuming the same food items, did not (Davis 1961).

Still, examples of large intestine length and weight changes attributed to changes in diet can be found in the literature. Seasonal variation in large intestine size in snow geese (Ankney 1977), gadwalls (Paulus 1982), and spruce grouse (Pendergast and Boag 1973) is usually explained by the seasonal variation in the amount of food eaten. However, other seasonal factors, photoperiod, temperature, etc., could be involved. Savory and Gentle (1976a, 1976b) were able to produce increases in Japanese quail large intestine length by adding fiber to the diet.

GENERAL METHODS

All birds used in this study were captured in the environs of Manhattan, Kansas (Netting locations, Appendix A) from November 1986 to February 1987. Birds were initially confined two per cage in either 46 x 22 x 27 cm or 38 x 22 x 27 cm cages. The cages were placed in a 3.2 x 3.8 x 3.2 m environmental chamber maintained at 10 C with a 10L:14D photoperiod. Relative humidity ranged from 76 to 78%.

Water and a maintenance diet of pelleted mash (label, Appendix B) were provided ad libitum. Mash was run through a 4.7 mm screen, and no grit was provided.

Evisceration and Measurement Technique

Bird body weights were recorded to 0.01 gm within 15 minutes of the start of the photoperiod, and also at the time of death. After death by cervical dislocation, wing length was measured to the nearest mm from the proximal tip of the carpometacarpus to the end of the leading primary. If possible, age was determined (adult or juvenile) by removing the skin from the top of the head and checking the degree of skull ossification (Wiseman 1962). Each bird was then examined externally for any signs of disease or injury, and prepared for evisceration.

After a ventral incision through the sternum was performed, sex was determined by gonadal inspection.

The gastro-intestinal tract and associated organs were then removed from the body cavity. The esophagus, proventriculus, and pancreas were all disjoined from the remainder of the tract and discarded. The small intestine was isolated by disjunctions at the gizzard and illeo-cecal junction. The ceca were disjoined from the small intestine, and the large intestine was considered to be the portion of the tract from the illeo-cecal junction up to and including the cloaca. All extraneous fat deposits were removed from the intestines and the gizzard with forceps.

All intestinal parts (small intestine, large intestine and ceca) were measured to 1.0 mm using Leopold's straight ruler technique (Freeling and Moore 1987). Cecal lengths were combined, and the ceca were weighed together. One individual measured all intestinal parts. Wet weights of the small intestine, large intestine, combined ceca, gizzard, and liver were recorded to 0.1 mg. All organs were patted dry prior to weighing, and gizzards were stripped of their contents.

After measuring and wet weighing, intestinal pieces were either placed in glass jars containing 50 ml 10% formalin acetate (label, Appendix C) for fixation and preservation, or immediately dried. If the intestinal parts were fixed, the liver and gizzard were frozen at -5 C for later drying. Otherwise, the liver and the

gizzard were dried immediately with the intestinal parts. All organs were dried at 65 C for 24 hours, and then reweighed to 0.1 mg.

Statistics

The significance level for all tests was $p=0.05$. Analyses were performed using t-tests, and one- and two-way analysis of variance (ANOVA) procedures. ANOVA for a 4 x 4 Latin Square design was also performed. Fisher's protected least significant difference (Ott 1984) was used for pairwise comparisons.

PROXIMATE ANALYSIS

Methods

The nutritive value of feed used in this study was estimated by proximate analysis as follows. Random samples of each diet were ground into homogenous units. Moisture content was determined by drying a sample to a constant weight and ascertaining the weight (water) loss. A dried sample was ignited in a furnace at approximately 600 C to obtain ash content. Crude fat was determined by ether extraction (Cullison 1982) and crude protein was analyzed indirectly by the Kjeldahl method (Perry 1984). The residue remaining after simulated acidic and alkaline digestion was the crude fiber portion of the total carbohydrates (Nagy and Haufler 1980). Nitrogen-free extract, the other usable carbohydrate portion, was determined indirectly by subtraction of the other 5 proximate analysis fractions from 100%. Gross energy was determined by bomb calorimetry.

Results

Sunflower seeds contained the highest energy content, highest protein content, least fiber, least nutrients, and the least moisture of the three feeds analyzed (Table 1). The balanced mash contained almost as much protein as the sunflower seeds, but had much less energy. The mash also had a large fiber and

Table 1. Selected nutritional characteristics of chick starter (balanced mash), white proso millet, and black oil sunflower seeds as determined by bomb calorimetry and proximate analysis.

	Mash	Millet	Sunflower
Gross Energy (kcal/g)	4.6	4.7	7.5
Crude Protein (%) ^A	24.9	11.4	25.7
Ether Extract (%)	1.6	4.2	56.1
Crude Fiber (%)	4.5	6.5	3.1
Ash (%)	6.8	3.8	3.3
Nitrogen-free extract (%)	62.2	74.1	11.8
Moisture (%)	12.5	12.9	6.3

^Aall percentages based on dry weight, except moisture.

mineral nutrient component. Millet was the poorest diet of the three analyzed, containing the most fiber and the least amount of protein.

EXPERIMENT I

POST MORTEM CHANGES IN DIGESTIVE ORGAN MORPHOLOGY

METHODS

Dark-eyed juncos (Junco hyemalis) were maintained in the laboratory under a 10L:14D photoperiod at 10 C for 14 days prior to experimentation. Five days prior to experimentation, the juncos were sorted into two groups according to wing length, skull ossification, plumage, and weight. Age was determined by skull ossification (Wiseman 1962) in conjunction with plumage characteristics (Blake 1964, Grant and Quay 1970). Wing length was used to distinguish adult males from adult females and from juveniles of either sex (Balph 1975, Blake 1967, Grant and Quay 1970, Wood 1969, Yunick 1981).

After sorting, adult male birds were inserted into a 4 x 4 Latin Square cage arrangement. Treatments consisted of evisceration at 0, 30, 60, and 90 minutes after death. Four birds were sacrificed at the start of the light period per day, and day effects were confounded with the row in which the bird was in the square. Digestive organ measurements were performed according to the procedures described previously.

Balanced mash and water were provided ad libitum during the experiment. Birds of unknown sex and age were inserted into the Latin Square cage matrix after

the removal of the adult males, and the experiment was repeated.

RESULTS

Cranial ossification in juveniles identified by wing length (Balph 1975, Blake 1967, Grant and Quay 1970, Wood 1969, Yunick 1981) and plumage (Blake 1964, Grant and Quay 1970) was complete in some individuals. Therefore, females could not be accurately aged.

Females had nearly identical mean wing lengths (75.1 mm) and body weights (17.8 g) as the juvenile males (75.3 mm, 17.8 g, respectively). Adult males were significantly heavier (19.1 g) and had larger wing lengths (80.6 g). Despite the differences in body size, the only statistically significant difference in the lengths and weights of the digestive organs of the three groups (adult males, juvenile males, and females) was the dry weight of the ceca (Table 2). Juvenile male ceca (1.6 mg) were smaller than both female (2.0 mg) and adult male (2.2 mg) ceca. Small intestine, large intestine and liver weights showed some variability between the three groups, while gizzard weights and small intestine lengths were nearly identical.

No significant differences in any of the morphological characteristics measured were found when the birds were eviscerated up to 90 minutes after death (Table 3). Cecal lengths increased gradually from 5.1

Table 2. Mean wing lengths (mm), body weights (g), and digestive organ weights (mg) and lengths (mm) from adult male (n=16), juvenile male (n=6), and unknown age female (n=10) dark-eyed juncos, all sampling times after death combined.

	Ad. Males	Juv. Males	Females	SE
Wing length	80.6 ^A	75.3 ^B	75.1 ^B	0.54
Body weight	19.1 ^A	17.8 ^B	17.8 ^B	0.40
Small intestine				
length	140.4	138.5	143.6	2.43
wet weight	601.9	635.0	677.0	34.95
dry weight	140.6	138.5	154.0	7.87
Large intestine				
length	14.4	12.6	12.9	1.16
wet weight	41.9	36.7	39.0	4.65
dry weight	5.8	5.3	5.4	0.59
Ceca (combined)				
length	5.3	5.8	6.2	0.39
dry weight	2.2 ^A	1.6 ^B	2.0 ^A	0.16
Gizzard				
wet weight	593.8	591.7	603.0	22.43
dry weight	169.4	171.9	180.1	7.94
Liver				
wet weight	715.0	776.7	703.0	35.16
dry weight	208.9	220.2	204.2	6.89

Means with the same letter are not significantly different between sex/age groups.

Table 3. Mean wing lengths (mm), body weights (g), and digestive organ lengths (mm) and weights (mg) from dark-eyed juncos eviscerated at selected times after death. (n=8)

	Time after death eviscerated (mins)				
	0	30	60	90	SE
Wing length	77.3	77.6	78.7	77.9	1.14
Body weight	18.7	18.1	18.0	19.0	0.52
Sm. Intestine					
length	142.1	140.2	138.5	143.3	2.97
wet weight	650.0	653.8	585.0	637.5	40.75
dry weight	159.7	147.5	129.1	141.3	9.25
Lg. Intestine					
length	13.6	12.0	13.2	15.1	1.23
wet weight	40.0	40.0	36.2	43.8	4.93
dry weight	6.8	5.4	4.7	5.4	0.50
Ceca					
length	5.1	5.8	5.3	6.5	0.46
dry weight	2.2	2.1	1.9	2.1	0.19
Gizzard					
wet weight	582.5	613.8	582.5	606.3	22.71
dry weight	174.6	175.8	165.9	176.5	8.42
Liver					
wet weight	637.5	757.5	750.0	746.3	36.33
dry weight	190.3	218.5	212.6	216.9	10.75

mm (0 minutes after death) to 6.5 mm (90 minutes after death), but the increase was not significant.

Cage position had no significant effect on digestive organ length and weight.

DISCUSSION

The negligible effect of the sex of dark-eyed juncos on digestive organ morphology is consistent with the lack of sex influenced digestive organ differences found in house sparrows (Passer domesticus) (Gier and Grounds 1944) and California quail (Leopold 1953). Different seasonal digestive organ morphologies depending on the sex of the bird were reported by Davis (1961) and Pendergast and Boag (1973) in rufous-sided towhees and spruce grouse respectively. Liver weights also may be sexually influenced, increasing in prebreeding females, possibly because of hyperphagia (Drobney 1984, Pulliainen and Tunkkari 1984). Sexes could likely be pooled for analysis, if the environmental demands operating on each sex were similar (ie. not during the breeding season).

Age also appeared to have little effect on the lengths and weights of the digestive organs. House sparrows (Gier and Grounds 1944) and California quail (Leopold 1953) also show no differences in digestive organ morphology due to age. Kirkpatrick (1944) found that both ceca and intestinal lengths stabilize in pheasants at approximately 80 days of age and do not change for a least 9 months thereafter, when the birds are fed the same diet. Age is usually not determined for digestive organ studies, and appears to have little

effect on digestive organ morphology except if the juveniles are extremely young.

The time eviscerated and measured after death (up to 90 minutes) had no significant influence on any morphological characteristics, indicating that it is not critical to measure gut morphology immediately after death if just length and weight measurements are desired. However, if histological observations are warranted, measurement should take place immediately after death to prevent autolysis and other artifacts from appearing (Fenwick 1982).

EXPERIMENT II

DIURNAL VARIATION IN DIGESTIVE ORGAN MORPHOLOGY

The objective of this experiment was to determine the effects, if any, of the time of day on digestive organ length and weight.

METHODS

Dark-eyed juncos were maintained in the laboratory under a 10L:14D photoperiod at 10C for 14 days prior to experimentation. The birds were randomly inserted into the 4 x 4 Latin Square cage arrangement and sampling grid. Experimental periods lasted four days each with four birds sacrificed per day, one each at 0, 3, 6, and 9 hours after the onset of the light period. Two types of trials, food deprivation and free feeding, were each performed twice (Table 4).

In food deprivation, food was removed from the cage at the start of the light phase of the photoperiod. In free-feeding trials, balanced mash was provided ad libitum, and water was available ad libitum in all trials.

Birds were weighed at the start of the light period and at the time of evisceration. During food deprivation trials, intestines were empty and did not need purging before being measured. During the feeding trials, intestines were cleaned of food contents by gentle scraping with a scalpel. While this undoubtedly

Table 4. Summary of Experiment II trial dates and the days birds were held in captivity prior to experimentation.

Trial	Type	Date	Days in Captivity
1	Food Deprived	16-19 Dec. 1986	19
2	Free-Feeding	26-29 Jan. 1987	14
3	Food Deprived	2- 5 Feb. 1987	21
4	Free-Feeding	9-12 Feb. 1987	28

removed some of the intestinal mucosa, such loss was considered negligible when compared to total intestinal weights. Weights of the intestines were taken both before and after purging.

Results

Food deprived birds showed a gradual decrease in body weight, losing approximately 0.20 g/hr during the 9 hours studied. Free-feeding juncos showed an abrupt increase in body weight after only 3 hours (1.61 g), and maintained that extra weight throughout the day (Table 5). Free-feeding birds were significantly heavier at all three sampling periods after the start of the light phase of the photoperiod, with a maximum weight difference of 3.9 g at the end of the 9 hours. Initial weights for both feeding regimes ranged from 18.19 to 18.68 g, with no significant differences. Wing lengths ranged from 75.6 to 79.1 mm the difference approached significance only at the third hour after the beginning of the photoperiod.

Small intestine length started out nearly the same in both food deprived birds (144.1 mm) and free-feeding birds (142.9), but quickly increased in the free-feeding juncos to 160.4 mm at 3 hours after the start of the photoperiod (Table 6). A maximum length of 170.3 mm was reached at the ninth hour. Small intestine length in food deprived birds decreased slightly after the start of the photoperiod to 139.8 mm, remained stable at 139.5 mm, and then decreased significantly to 135.3 mm.

Wet weights of the small intestine increased significantly in the free-feeding birds from 512.8 to

Table 5. Mean wing lengths (mm), body weights (g), and weight changes of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

	Hour After the Start of the Photoperiod				
	0	3	6	9	SE
<hr/>					
Wing length					
food deprived	77.5	79.1	79.0	78.8	0.97
free-feeding	76.9	75.6	77.3	77.0	
Initial weight					
food deprived	18.38	18.53	18.63	18.19	0.33
free-feeding	18.55	18.59	18.68	18.29	
Final weight					
food deprived		17.87 ^A	17.33 ^A	16.48 ^B	0.28
free-feeding		20.21 ^C	19.85 ^C	20.38 ^C	
Weight change					
food deprived		-0.65	-1.31 [*]	-1.72 [*]	
free-feeding		1.61 [*]	1.17	2.09 [*]	

^{*}Significant weight change.

Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.

Table 6. Mean lengths (mm) and weights (mg) of the small intestines of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

	Hour After the Start of the Photoperiod				
	0	3	6	9	SE
<hr/>					
Length					
food deprived	144.1 ^A	139.8 ^A	139.5 ^A	135.3 ^B	2.98
free-feeding	142.9 ^A	160.4 ^C	157.6 ^C	170.3 ^D	
Wet weight					
food deprived	608.7 ^A	624.4 ^A	542.7 ^A	501.9 ^A	64.77
fed(-contents)	512.8 ^A	553.5 ^A	494.9 ^A	562.4 ^A	
fed(+contents)	512.8 ^A	1035.3 ^B	938.1 ^B	1101.7 ^B	
Dry weight					
food deprived	158.1	160.1	127.1	129.5	13.01
free-feeding	128.7	162.5	151.0	176.6	

Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.

1035.3 mg after 3 hours, and then did not change significantly. When the contents were removed from the intestines of free-feeding birds, wet weight decreased to 553.5 mg and the statistically significant difference disappeared. Small intestine wet weights did not differ between food deprived and free-feeding juncos if the intestines were purged. Dry weights mimicked wet weights.

Large intestine lengths (Table 10) did not vary significantly throughout the treatments, but increased slightly from 9.3 to 11.1 mm in the free-feeding birds. A significant interaction between the time of day and feeding regime was detected with large intestine wet weight, and became more pronounced when the large intestine was dried. Dry weight increased in the free-feeding birds from 4.8 mg at the start of the photoperiod to 7.2 mg at the last hour sampled, while at the same time decreasing from 6.1 to 4.3 mg in the food deprived birds over the same time span.

A high degree of variability in large intestine wet and dry weights was evident. Wet weights between the two feeding regimes differed by 30% at the start of the photoperiod when they should have been nearly equal. Dry weights reflected wet weights, with food deprived birds having 1.3 mg heavier large intestine dry weights than free-feeding birds at a time when they should have

Table 7. Mean lengths (mm) and weights (mg) of the large intestines of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

	Hour After the Start of the Photoperiod				
	0	3	6	9	SE
<hr/>					
Length					
food deprived	8.5	9.0	8.5	8.8	0.77
free-feeding	9.3	10.8	9.3	11.1	
Wet weight					
food deprived	27.2	25.4	21.5	21.0	2.19
free-feeding	20.8	25.6	18.7	23.2	
Dry weight					
food deprived	6.1 ^{AB}	6.2 ^{AB}	5.5 ^{BC}	4.3 ^C	0.55
free-feeding	4.8 ^{BC}	7.3 ^A	6.3 ^{AB}	7.2 ^A	

Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.

been equal.

Cecal lengths and weights also showed much variation, with birds at the start of the photoperiod from the free-feeding trials having longer (6.0 to 5.5 mm) and heavier wet weights (6.4 to 5.3 mg) and dry weights (1.8 to 1.3 mg) than the food deprived trial birds at the same time period (Table 8). Both groups should have been nearly equivalent at that time. Cecal dry weights are statistically significantly different, but any biological significance is masked by the significant difference in initial dry weights (1.8 to 1.3 mg).

Gizzard wet weights showed no significant differences, remaining near 600 mg throughout the time period sampled for the free-feeding birds and only slightly decreasing to 580 mg at the ninth hour sampled in the food deprived birds (Table 9). Dry gizzard weights also were not significantly different over the time frame studied in both groups of birds.

Wet liver weights significantly decreased from a high of 708.3 mg at the third hour after the start of the light phase to a low of 553.1 mg at the end of the sampling period in the food deprived birds. Free-feeding bird wet liver weights were significantly higher than those of the food deprived birds at each hour sampled after the start of the photoperiod,

Table 8. Mean lengths (mm) and weights (mg) of the ceca (combined) of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

	Hour After the Start of the Photoperiod				
	0	3	6	9	SE
<hr/>					
Length					
food deprived	5.5	5.3	6.0	5.6	0.50
free-feeding	6.0	6.8	6.6	6.1	
Wet weight					
food deprived	5.3	5.8	4.6	5.0	0.75
free-feeding	6.4	6.5	6.6	5.3	
Dry weight					
food deprived	1.3 ^A	1.6 ^{ABC}	1.5 ^{AB}	1.3 ^A	0.14
free-feeding	1.8 ^{BC}	1.8 ^{BC}	1.9 ^C	1.6 ^C	

Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.

Table 9. Mean gizzard and liver weights (mg) of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

		Hour After the Start of the Photoperiod				
		0	3	6	9	SE
		<hr/>	<hr/>	<hr/>	<hr/>	<hr/>
<u>Gizzard</u>						
Wet weight						
food deprived	640.1	588.5	572.4	580.0	23.93	
free-feeding	599.1	591.7	612.8	601.4		
Dry weight						
food deprived	181.2	169.2	166.8	171.2	7.18	
free-feeding	171.9	163.0	172.5	174.1		
<u>Liver</u>						
Wet weight						
food deprived	655.1 ^{AB}	708.3 ^A	569.2 ^{BC}	553.1 ^C	38.63	
free-feeding	684.0 ^{AB}	890.7 ^D	920.2 ^D	1054.4 ^E		
Dry weight						
food deprived	204.5 ^{AB}	217.3 ^A	177.0 ^B	166.4 ^B	13.07	
free-feeding	211.7 ^A	285.8 ^C	327.9 ^D	382.2 ^E		
Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.						

significantly increasing from 890.7 mg after 3 hours to 1045.4 at 9 hours. Dry liver weights were more sensitive to changes in feeding, showing significant increases in the free-feeding birds at each sampling point. At the start of the photoperiod, dry liver weights in the free-feeding birds were 211.7 mg, and then significantly increased after 3 hours to 285.8 mg, after 6 hours to 327.9 mg and after 9 hours to 382.2 mg. Dry liver weights in the food deprived birds differed significantly from the free-feeding birds at each hour after the start of the photoperiod, but differed from each other only after 6 hours when a 150 mg decrease occurred.

Wet and dry weights of the total digestive tract (gizzard, both intestines and ceca) showed no significant differences when each treatment was analyzed (Table 10). Total wet weights did increase slightly in the free-feeding birds from 1139.3 mg at the start of the photoperiod to 1192.2 mg after 9 hours; they also decreased in the food deprived birds from 1281.3 mg at the start of the photoperiod to 1107.8 mg after 9 hours. All values were within the range of normal responses suggested by the weights at the start of the photoperiod. Total dry weights also increased in the free-feeding birds (from 307.2 to 359.3 mg after 9 hours) and decreased in the food deprived birds (346.7

Table 10. Mean total digestive tract (gizzard, both intestines and ceca) weights (mg) and mean total intestinal (both intestines, with and without ceca) lengths (mm) of dark-eyed juncos either deprived of food or fed balanced mash and eviscerated at different times after the start of the photoperiod. (n=8)

		Hour After the Start of the Photoperiod				
		0	3	6	9	SE
Wet weight						
food deprived	1281.3	1244.1	1141.3	1107.8	48.45	
free-feeding	1139.3	1177.2	1132.9	1192.2		
Dry weight						
food deprived	346.7	337.2	300.9	306.2	15.50	
free-feeding	307.2	334.5	331.5	359.3		
Length						
food deprived	152.6 ^A	148.8 ^A	148.0 ^A	144.0 ^A	3.29	
free-feeding	152.1 ^A	171.1 ^B	166.9 ^B	181.4 ^C		
Dry weight						
food deprived	158.1 ^A	154.0 ^A	154.0 ^A	149.6 ^A	3.46	
free-feeding	158.1 ^A	177.9 ^{BC}	173.1 ^B	187.5 ^C		

Means unlettered or with the same letter (compared both horizontally between hours and vertically between feeding regimes) are not significantly different.

to 306.2 mg), but such variance was in the range suggested by the starting measurements before the trials began.

Total intestinal length without ceca did not significantly change from 152.6 mm at the start of the photoperiod to 144.0 mm after 9 hours in the food deprived birds, but significantly increased after 3 hours (152.1 mm up to 171.1 mm) and again at 9 hours (181.4 mm) in the free-feeding birds. Including cecal length caused the increase at 9 hours in the free-feeding birds to be less prominent.

DISCUSSION

The significant impact of time of collection and feeding upon small intestine length could explain the attributed dietary induced intestinal length changes reported for rufous-sided towhees (Davis 1961), mallards (Miller 1975), snow geese (Ankney 1977), Canada geese (Raveling 1979), wood ducks (Drobney 1984), and Lagopus spp. (Moss 1972, 1974, Pendergast and Boag 1971, 1973, Thomas 1984, Pulliainen and Tunkarri 1983). None of these authors reported the time of the day the birds were collected however, thereby making critical analysis of their work with respect to the observed diurnal variation in small intestine length somewhat speculative.

Lengthening of intestinal length in the winter is

attributed to increases in dietary fiber (switches to low energy vegetative matter when high energy insects are no longer available) which causes increased food consumption and subsequent hypertrophy of the small intestine (Pendergast and Boag 1973, Moss 1974, Ankney 1977, Raveling 1979, Pulliainen and Tunkarri 1983, Thomas 1984, Drobney 1984). This increase could be explained by the time available for foraging and collection time. Shorter, colder winter days result in increased food consumption in a condensed time frame compared to the summer months (West and Meng 1965), which would result in increased food being sent to the intestine. Intestines containing more food are longer, as shown by the increase in small intestine length during the photoperiod studied. The increased intestinal length is not due to hypertrophy of the intestinal tissue then, but merely a result of putting more food in the intestine (intestinal stretching) as shown by this experiment and hyperphagia studies (Haeger et a. 1953, Levin 1965, 1969). Since intestinal weights did not change during this experiment, it is unlikely that additional intestinal tissue was created. Therefore increased luminal nutrition (and subsequent increases in intestinal tissue) could not explain the quick lengthening of the intestine as suggested by Levin (1969).

Miller (1974) considers the ability of waterfowl to modify their gut length and weight a successful evolutionary strategy to utilize different food resources when required. While he did not mention his collection times, he did not remove intestinal contents prior to weighing. Miller's emphasis on the role of small intestine length could be well placed, but it is likely that the reason small intestines change in length is due to the amount of material in them which causes temporary stretching. While the limits of the intestine to stretch could be evolutionarily important, it is likely that the importance placed on small intestine length as a measure of digestibility (Moss 1977), fitness (Miller 1975), or diet quality (Leopold 1953, Moss 1974, Kehoe and Ankney 1985) is not well grounded.

Some of the discrepancies in the literature could be due to collection time. The lack of differences in small intestine length with food changes reported by Hupp and Brown (1984) and Kirkpatrick (1944), in sage grouse and ring-necked pheasants, respectively, could be explained if all birds were collected at the same time/hour of the day. However, this information is not given.

Diurnal variation in large intestine and cecal morphology may have occurred, but it was masked by the

high degree of variability within the control birds sacrificed at the start of the photoperiod. This variability could have been due to the low weights of the large intestine and ceca which could be highly influenced by small differences in disjunction location or fat removal. Moss (1983) experienced high variability in grouse small intestine lengths also.

The lack of daily fluctuation in gizzard weights, regardless of the food intake, does not discredit reported winter gizzard weight increases in gadwalls (Paulus 1982), wood ducks (Drobney 1984), Lagopus spp (Pendergast and Boag 1973, Hanssen 1979a), Canada geese (Raveling 1979), and sage grouse (Hupp and Brown 1984) attributed to increases in dietary fiber.

Liver weight was highly sensitive to food intake and time sampled, increasing in the free-feeding birds and decreasing in the food deprived ones. Liver weight increases reported for sage grouse (Hupp and Brown 1984), wood ducks (Drobney 1984), snow geese (Ankney 1977) and Lagopus spp. (Thomas and Popko 1981, Pulliainen and Tunkkari 1984) during the spring should be viewed with some caution, since collection times are not given. Pulliainen (1985) also suggests caution in the interpretation of interspecific liver weights.

EXPERIMENT III

DIET AND GRIT INFLUENCES ON DIGESTIVE ORGAN MORPHOLOGY

The objective of this experiment was to attempt to modify the digestive organ morphology of Harris' sparrows by using different diet and grit regimes.

METHODS

Harris' sparrows were maintained in the laboratory using balanced mash and a 10L:14D photoperiod at 10 C for 30 days prior to experimentation. Nine birds per day for 5 days (45 birds total) were randomly assigned treatments of either control (balanced mash), white proso millet or black oil sunflower diets for periods of 7 and 14 days. Additionally, two birds per day on the 14 day millet or sunflower diet received commercial granite grit. One bird of the nine was immediately sacrificed, eviscerated, and measured (as stated in the GENERAL METHODS) at the start of the photoperiod.

Birds were initially given either 30 gm millet, 20 gm control, or 15 g sunflower, and then an additional 10 g millet, 15 g control, or 10 g sunflower during the remainder of the experiment. If receiving grit, 15 g of commercial granite grit was provided at the onset of the experiment. Water and food were available ad libitum. Two ml each of water soluble vitamins (Appendix D) and minerals (Appendix E) were included in the water daily.

Cages were placed in 48 x 25 x 13 cm polypropylene quail containers to capture spilled food. Spilled food and excreta were separated, oven-dried, and then subtracted from the amount provided to determine the amount consumed.

Collected data were analyzed to determine the influences of 1) diet and 2) grit on the length and weights of the digestive tract organs.

PART A - DIETARY INFLUENCES ON HARRIS' SPARROW DIGESTIVE

ORGAN MORPHOLOGY

RESULTS

No differences were significant for initial weights, final weights or wing length between the individual treatments, although when all sampling days were combined the final weights of birds consuming millet (32.74 g) were significantly less than the sunflower (34.87 g) and mash (34.89 g) eating birds (Table 11). With all days combined, the sparrows consuming millet significantly lost weight (-1.43 g), the sparrows consuming mash gained weight (0.59 g) and the sparrows consuming sunflower maintained a steady weight. The birds consuming mash for the entire 14 days were the only individual treatment to show a statistically significant weight increase (0.91 g) (Table 12).

Food consumption was significantly different among day 7 birds with 12.02 g/bird/day of mash, 7.23 g/bird/day of millet, and 5.37 g/bird/day of sunflower being consumed. Mash consumption significantly declined down to 7.92 g/bird/day at day 14, and was no longer different from millet consumption (6.78 g/bird/day). Overall, 9.97 g/bird/day of mash, 7.00 g/bird/day of millet, and 4.92 g/bird/day of sunflower were eaten.

Small intestine length was not significantly

Table 11. Mean wing lengths (mm), body weights (g), total digestive tract (gizzard, both intestines, and ceca) weights (mg) and total intestinal (both intestines, with and without ceca) length (mm) from Harris' sparrows fed three diets, all days combined.

	Diet			SE
	Millet	Mash	Sunflower	
Number	10	10	10	
Wing length	87.5	87.7 ⁺	88.0	1.14
Initial wt.	34.17	34.30	34.93	0.84
Final wt.	32.74 ^A	34.89 ^B	34.87 ^B	0.78
Wt. change	-1.43 [*]	0.59 [*]	0.06	
Food consumed/ bird/day	7.00 ^B	9.97 ^A	4.92 ^C	0.46
Number	9	14	10	
Tot. wet wt.	1945.8 ^A	2073.0 ^A	2276.1 ^B	65.63
Tot. dry wt.	364.7 ^A	411.8 ^B	433.7 ^B	12.06
Tot. len.	185.0	187.0	189.0	4.20
Tot. len.(+ceca)	191.8	193.9	195.6	4.27

⁺n=15.

^{*}significant weight change.

Means with the same letter (compared horizontally) are not significantly different.

Table 12. Mean wing lengths (mm), body weights (g), and daily food intake (g), from Harris' sparrows fed three food types over 14 days. (n=5)

	Diet			
	Millet	Mash	Sunflower	SE
Wing length				
Day 0		88.2		
Day 7	88.6	88.2	88.8	1.66
Day 14	86.4	86.6	87.2	
Initial wt.				
Day 0		34.92		
Day 7	34.71	35.19	35.12	1.22
Day 14	33.63	32.27	34.73	
Final wt.				
Day 7	34.05	36.05	35.29	
Day 14	31.43	33.68	34.44	1.08
Wt. change				
Day 7	-0.66	0.86	0.17	
Day 14	-2.20*	0.91*	-0.29	
Food consumed/ bird/day				
Day 7	7.23 ^A	12.02 ^B	5.37 ^C	
Day 14	6.78 ^A	7.92 ^A	4.48 ^B	0.46

*significant weight change.
Means with the same letter (compared both horizontally between diets and vertically between days) are not significantly different.

influenced by diet, although mean length for 14 day sunflower consuming birds was 11 mm longer than the day 0 control birds (Table 13). Wet weights of the small intestines were heavier in all birds, except the 14 day millet group, than the 966.9 mg weight of the control group sacrificed immediately. A high degree of variability (Standard Errors were 5 to 10% of the mean values) kept any of the individual treatment wet and dry weight differences from achieving significance. With all days combined, small intestine wet weights from the birds on a sunflower diet were significantly higher than those from the birds consuming millet (1127.2 to 970.1 g) (Table 14). Dry small intestine weights were not significantly different between the diets however.

Sparrows consuming millet had longer large intestines (day 7 = 15.8 mm, day 14 = 15.3 mm) than the sunflower eating birds (day 7 = 14.0 mm, day 14 = 14.4), although the difference was not statistically significant (Table 15). Mash consuming birds showed extreme variability in large intestine length, being 14.0 mm on day 0, increasing to 15.5 mm on day 7, and then decreasing back to 14.2 mm on day 14. Wet and dry weights again showed a high degree of variability, with standard errors over 10% of the mean values.

Combined cecal lengths remained fairly constant, with no appreciable differences from the 6.8 mm value

Table 13. Mean small intestine weights (mg) and lengths (mm) from Harris' sparrows fed three diets over 14 days. (n=5)

Day	Diet	Length	Wet Weight	Dry Weight
0	Mash	170.0	966.9	126.6
	Mash	177.8	1097.2	114.3
7	Millet	173.0	1025.1	107.4
	Sunflower	168.6	1100.2	101.2
	Mash	172.4	1082.0	114.2
14	Millet	164.6	915.1	98.6
	Sunflower	181.0	1154.1	107.5
Standard Error		5.00	57.45	8.35

Table 14. Mean digestive organ weights (mg) and lengths (mm) from Harris' sparrows fed three food types, all sampling days combined.

	Diet			SE
	Millet	Mash	Sunflower	
	n=10	n=15	n=10	
Small intestine				
length	168.8	173.4	174.8	3.79
wet weight	970.1 ^A	1048.7 ^{AB}	1127.2 ^B	39.14
dry weight	103.0	118.4	104.2	5.62
Large intestine				
length	15.6 [#]	14.5 ⁺	14.2	0.90
wet weight	47.6 [#]	42.9 ⁺	51.0	3.78
dry weight	6.0 [#]	6.0 ⁺	6.6	0.43
Ceca (combined)				
length	6.8	6.9	6.6	0.24
wet weight	4.6	5.0	4.8	0.55
dry weight	1.4	1.4	1.4	0.12
Gizzard				
wet weight	908.6 ^A	978.6 ^A	1093.1 ^B	29.04
dry weight	264.5 ^A	283.4 ^A	321.5 ^B	8.54
Liver				
wet weight	1063.6 ^A	1283.7 ^B	974.4 ^A	60.10
dry weight	330.8 ^A	394.3 ^B	298.8 ^A	16.45

[#]n=9.

⁺n=14.

Means with the same letter are not significantly different between diets.

Table 15. Mean weights (mg) and lengths (mm) of the large intestines and combined ceca from Harris' sparrows fed three diets over 14 days. (n=5)

Day	Diet	Length	Wet Weight	Dry Weight
<u>Large Intestine</u>				
0	Mash	14.0	44.4	6.6
7	Mash ⁺	15.5	53.1	6.7
	Millet	15.8	48.1	6.0
	Sunflower	14.0	44.3	6.1
14	Mash	14.2	43.3	4.9
	Millet ⁺	15.3	46.9	6.0
	Sunflower	14.4	57.7	7.0
Standard Error		1.39	5.26	0.59
<u>Ceca</u>				
0	Mash	6.8	5.1	1.5
7	Mash	6.5	5.2	1.4
	Millet	6.6	5.1	1.6
	Sunflower	6.6	5.2	1.4
14	Mash	7.2	4.7	1.3
	Millet	7.0	4.2	1.3
	Sunflower	6.6	4.5	1.3
Standard Error		0.34	0.82	0.16
⁺ n=4.				

of the control birds at day 0. Cecal wet weight dropped slightly in the birds fed for 14 days (mash = 4.7 mg, millet = 4.2 mg, sunflower = 4.5 mg) when compared to the day 0 and day 7 birds (day 0 = 5.1 mg, day 7 mash = 5.2 mg, millet = 5.1 mg, sunflower = 5.2 mg) but the decrease was not significant. Cecal dry weights were 6 to 7% lower in the birds fed for 14 days in relation to all other groups.

Gizzard weights varied significantly between the three diets over the 14 days studied (Table 16). Sunflower consumption produced the heaviest gizzards (wet weight 1051.4 mg on day 7 and 1134.7 mg on day 14), while millet consumption produced the lightest ones (wet weight 890.2 mg on day 7 and 927.1 mg on day 14). Mash produced gizzards ranging from 949.5 mg to 1016.4 mg wet weight. Dry weight mimicked wet weight. Although gizzard weight did not significantly change from day to day, a general increase for all three diets occurred from day 7 to day 14.

Liver wet weights remained unchanged in the birds on a mash diet over 14 days (day 0 = 1209.4 mg, day 7 = 1301.1 mg, day 14 = 1340.7 mg) while decreasing in the birds consuming millet (day 7 = 1177.6 mg, day 14 = 949.4 mg), and sunflower (day 7 = 875.3 mg, day 14 = 1073.3 mg). Dry weights again merely reflected wet weights. A significant interaction between the day

Table 16. Mean wet and dry weights (mg) of gizzards and livers from Harris' sparrows fed three diets over 14 days. (n=5)

Day	Diet	Gizzard		Liver	
		Wet Wt.	Dry Wt.	Wet Wt.	Dry Wt.
0	Mash	969.9 ^B	283.6 ^B	1209.4 ^{AB}	369.5 ^{AB}
	Mash	949.5 ^{AB}	274.1 ^{AB}	1301.1 ^{AB}	399.1 ^B
7	Millet	890.2 ^A	262.1 ^A	1177.6 ^{ABC}	362.1 ^{ABC}
	Sunflower	1054.1 ^{BC}	316.3 ^{BC}	875.3 ^D	271.9 ^D
14	Mash	1016.4 ^{BC}	292.4 ^{BC}	1340.7 ^A	414.3 ^A
	Millet	927.1 ^{AB}	267.0 ^{AB}	949.4 ^{CD}	299.4 ^{CD}
	Sunflower	1134.7 ^C	326.8 ^C	1073.3 ^{BCD}	324.8 ^{BCD}
Standard Error		39.62	10.78	82.01	22.26

Means with the same letter vertically are not significantly different.

sampled and diet consumed occurred with liver weight, with the liver weight of birds on either mash or sunflower diets increasing throughout the experiment. Liver weights in the birds consuming millet decreased from day 7 to day 14.

Total digestive tract wet weights for each of the diets at each day sampled were heaviest in the birds consuming a sunflower diet (2201.1 mg at day 7 and 2351.0 mg at day 14) (Table 17). Millet consumption produced the lightest tracts (1968.6 mg at day 7 and 1917.3 mg at day 14) and mash consumption produced mean tract weights from 1976.3 mg to 2146.4 mg. None of the means is significantly different unless the days are pooled. Total tract dry weights exhibited smaller respective differences between treatments than did total tract wet weights.

Total intestinal length, both with and without ceca included, was not significantly different between treatments.

Table 17. Mean wet and dry weights (mg) of the total digestive tract (gizzard, both intestines, and ceca) and mean total intestinal (both intestines, with and without ceca) length (mm) from Harris' sparrows fed three diets over 14 days. (n=5)

Day	Diet	Total Tract		Total Int. Length	
		Wet Wt.	Dry Wt.	W/O Ceca	W/ Ceca
0	Mash	1976.3	418.4	184.0	290.8
	Mash	2102.4	402.2	191.3	197.8
7	Millet	1968.6	377.1	188.8	195.4
	Sunflower	2201.1	424.8	182.6	189.2
	Mash	2146.4	412.8	186.6	193.8
14	Millet	1917.3	371.3	180.3	187.3
	Sunflower	2351.0	442.6	195.4	202.0
Standard Error		96.74	17.31	5.73	5.81

DISCUSSION

Food consumption data for sunflower and millet agree closely with that collected by Schuman (1984), who enclosed the entire cage with screen. Using the available quail cages made the sparrow cages easily accessible (unlike the screen covering) and also provided fairly precise results. Mash consumption is probably inflated at day 7 due to inexperience in separating mash from fecal material, and day 14 likely provides a better estimate of mash consumption.

If a high fiber diet produces long small intestines as hypothesized by Leopold (1953), Moss (1974), Miller (1975) and several other authors, then the birds consuming millet should have had significantly longer small intestines than the sunflower consuming birds. This did not occur though. Only Hupp and Brown (1984) and Kirkpatrick (1944) could find no dietary influences on small intestine length (using sage grouse and ring-necked pheasants), reporting similar results to those obtained in this experiment.

Experimental studies using Japanese quail (Savory and Gentle 1976b) and semi-domesticated mallards (Miller 1975) produced longer small intestines by either adding extra cellulose to the diet, or feeding high fiber plant material. It is difficult to evaluate these studies though, since the evisceration times were not thought to

be important and were not published.

The lack of any detectable difference in small intestine length in this experiment could be due to the sampling of the birds when their intestines were empty, since earlier research in this thesis clearly shows how the amount of food in the digestive tract can influence small intestine length. If the Harris' sparrows were allowed to forage on their respective diets and sampled later in the day, the high fiber millet by its bulk may have increased small intestine length over the low fiber, low consumption sunflower diet.

Wet and dry weights of both intestines and ceca did not vary depending on diet, probably since they were empty when measured. Small intestine wet weight differences after emptying of the contents were found interspecifically by Thomas (1984) and intraspecifically by Pendergast and Boag (1973). Other studies (Miller 1975, Savory and Gentle 1976a, 1976b) found significant small intestine wet weight differences among birds fed different diets, but they did not empty intestinal contents prior to weighing.

Large intestine length was not very sensitive to dietary changes, as Leopold (1953) and Miller (1975) found, and in contradiction to Ziswiler and Farners' contention that the large intestine is the most sensitive of all the digestive organs to dietary

influences. The seasonal variation in large intestine length reported in snow geese (Ankney 1977), gadwalls (Paulus 1982), and spruce grouse (Pendergast and Boag 1973) could have been due to sampling time inconsistencies.

Cecal length also was unaffected by diet in Harris' sparrows, contrary to studies published about waterfowl by Miller (1975), Paulus (1982), Drobney (1984), and Halse (1984) and about gallinaceous birds by Leopold (1953), Lewin (1963), Moss (1972), Pendergast and Boag (1973), Savory and Gentle (1976b), Gasaway (1976a), Pulliainen and Tunkkari (1983), and Thomas (1984). Hupp and Brown (1984) found no changes in spruce grouse ceca length in the spring though. The lack of effect of diet on cecal length could be due to the reduced function of the ceca in most passeriform birds (Ziswiler and Farner 1972), or could have been due to sampling when the ceca were empty. It is not know when ceca were measured in all of the other studies.

Differences in gizzard weight, though not significant, were contrary to that expected from the literature. Gizzard weight usually increases with increasing dietary fiber (Spitzer 1972, Pendergast and Boag 1973, Miller 1975, Savory and Gentle 1976b, Hanssen 1979a, Raveling 1979, Paulus 1982, Drobney 1984, Herd and Dawson 1984 , Hupp and Brown 1984, Kehoe and Ankney

1985, Hobaugh 1985). Sunflower may have been harder for the gizzard to grind than the higher fiber millet, and thus may have produced larger gizzards. The texture of the diet influences gizzard morphology (Ziswiler and Farner 1972).

A negative relationship between dietary fiber and liver weight (Drobney 1984, Pulliainen and Tunkkari 1984) was not evident with Harris' sparrows on three feeds. The results were more in line with lack of change in liver weights with different diets reported by Thomas et. al. (1975) and Pendergast and Boag (1971). The decline of liver weights in all but the mash consuming birds could have been due to a reduction in stress as discovered by Oakeson (1953, 1956) in white-crowned sparrows, but the sparrows should have been used to the mash diet after feeding on it for more than 30 days.

Total tract (gizzard, both intestines and ceca) weights were lowest in the birds consuming millet, just the opposite of Leopold's (1953), Moss' (1974), and Miller's (1975) results. The above mentioned authors did not mention removing intestinal contents prior to weighing, which may have influenced the results they obtained. The decrease in total tract weight in millet was influenced in this study by the weight loss experienced by those birds consuming millet. The weight

stable sunflower and mash consuming birds showed no tract weight differences.

Whether the lack of differences in intestinal length and most other organ measurements is due to sampling time or is a species specific phenomenon of Harris' sparrows could not be determined.

PART B - GRIT INFLUENCES ON HARRIS' SPARROW DIGESTIVE

ORGAN MORPHOLOGY

RESULTS

Only those sparrows consuming millet without grit showed a significant weight loss (-2.20 g) although mean weights decreased in all treatments (Table 18). Eviscerated weights were lowest in the millet (w/o grit) (31.43 g) and sunflower (w/grit) (31.61 g) treatments, respectively, due to lower starting weights and weight losses. Sunflower (w/o grit) and millet (w/grit) final weights were 34.44 and 34.90 g respectively. Birds on a millet diet consumed at least 2 g more of feed/day than did the birds consuming sunflower, regardless of grit, but were much better able to maintain body weight on millet if grit was available.

Small intestine length was not affected by either diet or grit (Table 19). Small intestine wet weight was significantly smaller in the millet (w/o grit) (915.1 mg) than both the millet (w/grit) (1053.4 mg) and sunflower (w/o grit) (1154.1 g) treatments. A significant interaction between the diet consumed and the presence of grit was detected for small intestine wet weight. Dry weight differences were not significant.

No large intestine or cecal lengths or weights were significantly different. However, cecal wet weights

Table 18. Mean wing lengths (mm), body weights (g) and daily food intake of Harris' sparrows fed millet and sunflower seeds with and without grit. (n=5)

	Millet		Sunflower		SE
	W/O Grit	W/ Grit	W/O Grit	W/ Grit	
Wing length	86.4	88.2	87.2	86.2	1.82
Init. weight	33.63	35.40	34.73	32.11	1.16
Final weight	31.43 ^A	35.90 ^B	34.44 ^B	31.61 ^A	1.02
Weight change	-2.20 [*]	-0.50	-0.29	-0.49	0.84
Food consumed/ bird/day	6.78 ^A	6.24 ^A	4.48 ^B	3.97 ^B	0.46

*Significant weight change.
Means with the same letter horizontally are not significantly different.

Table 19. Mean lengths (mm) and weights (mg) of the intestines and ceca from Harris' sparrows on two diets, with and without grit. (n=5)

	Millet		Sunflower		
	W/O Grit	W/ Grit	W/O Grit	W/ Grit	SE
Sm. intestine					
Length	164.6	183.2	181.0	170.6	4.66
Wet weight	915.1 ^A	1053.4 ^B	1154.1 ^B	1003.5 ^{AB}	43.85
Dry weight	98.6	102.8	107.5	94.5	6.71
Lg. intestine					
Length	15.3	13.6	14.4	15.0	0.99
Wet weight	46.9	51.9	57.7	49.0	4.70
Dry weight	6.0	6.2	7.0	6.4	0.56
Ceca (combined)					
Length	7.0	6.8	6.6	7.0	0.35
Wet weight	4.2	5.7	4.5	5.7	0.42
Dry weight	1.3	1.6	1.3	1.5	0.09
Means with the same letter are not significantly different.					

increased from means of 4.2 to 5.7 mg in the millet birds and 4.5 to 5.7 mg in the sunflower birds when grit was available. Dry cecal weight also increased from 1.3 to 1.6 (millet) and 1.5 (sunflower) mg in the birds which had access to grit.

Gizzard morphology was not affected by grit, and liver wet weights also showed no significant differences between the treatments (Table 20). However, liver dry weights were significantly higher in the birds consuming sunflower with no grit (369.9 mg) than the other three treatments. A significant interaction between the diet consumed and grit occurred with liver dry weight.

Total tract weight and intestinal length reflected the final weight pattern reported earlier.

Table 20. Mean gizzard, liver and total digestive tract (gizzard, both intestines and ceca) weights (mg) and intestinal lengths (both intestines, with and without ceca) from Harris' sparrows on two diets, with and without grit. (n=5)

	Millet		Sunflower		
	W/O Grit	W/ Grit	W/O Grit	W/ Grit	SE
Gizzard					
Wet weight	927.1	1041.7	1134.7	1018.0	74.65
Dry weight	267.0	302.1	326.8	294.7	20.70
Liver					
Wet weight	949.4	1209.4	1073.3	954.8	68.74
Dry weight	299.4 ^A	324.8 ^A	369.9 ^B	287.8 ^A	18.79
Total tract					
Wet weight	1917.3	2152.7	2351.0	2076.2	113.72
Dry weight	317.7	412.7	442.6	397.1	25.05
Intestinal					
Length	180.3	196.8	195.4	185.6	4.67
Len. (+ceca)	187.3	203.6	202.0	192.6	4.96

Means with the same letter are not significantly different.

DISCUSSION

Millet was not a suitable diet for weight maintenance in Harris' sparrows, unless grit was provided. Nestler (1946) could find no difference in the survival of both young and adult bobwhites regardless of if grit was available. It is unlikely that Harris' sparrows recieved any additional mineral benefits from the grit, as hypothesized by Robel and Bisset (1979), because of the water soluble mineral and vitamin supplements given to all of the birds. Sadler (1961) and Verbeek (1971) did find mineral benefits from grit consumption, but this occurred during egg laying when calcium was required.

Spitzer (1972) reported that gizzards became more muscular and contained grit during dietary shifts from insects to seeds. Whether the grit, the diet change, or a combination of the two factors caused the increased gizzard change is unknown. Grit did not produce heavier gizzards in this study, suggesting that perhaps a more severe dietary change (instead of the 3% fiber difference between millet and sunflower) may be necessary to produce changes in gizzard morphology.

No other studies have been performed that examine the effects of grit on other digestive organs than the gizzard. The possible grit influences on small intestine and liver weight reported in this study were

likely due to the final weights of the birds in the different treatments. Cecal weight increases with the presence of grit for both diets may warrant further investigation.

OVERALL DISCUSSION

While the time measured after death has little effect on digestive organ length and weight, the time of day collected (depending on feeding pattern) has a significant influence on small intestine length and liver weight. No effect of time collected was observed for the any other organ measurement taken.

This influence of collection time is probably the reason why Harris' sparrow small intestines did not appear to be influenced by diet, unlike the experimental results obtained for mallards (Miller 1975) and red grouse (Moss 1972). While these authors noted a positive relationship between dietary fiber and digestive organ length and weight, they did not mention collection times. If the birds were allowed to forage prior to evisceration, the intestines containing the most food (a high fiber diet requires more food for the same amount of energy) would appear to be longer due to stretching. However, if the birds were eviscerated prior to foraging, all intestinal lengths should be the same (as was noted with the Harris' sparrows). Another possibility is that Harris' sparrows lack the ability to morphologically modify their gut with dietary changes.

Miller (1975) and Moss (1972) noted increases in cecal length with increasing dietary fiber also. However, cecal morphology was not affected by the time

of day (feeding) in dark-eyed juncos, possibly reflecting the hypothesized reduced role of the ceca in this species (Ziswiler and Farner 1972). The reduced role of the ceca could explain the lack of size difference in Harris' sparrows on different diets.

Miller (1975) also found small intestines to be heavier on a high fiber diet, but he did not remove intestinal contents prior to weighing. As the time of day experiment showed, even though intestinal weights increase during the day, the increase is merely due an increase in the intestinal contents, which is likely what happened in Miller's study.

Organ measurements taken during field studies also need to be evaluated by looking at collection times. The field observations of Leopold (1953), Davis (1961), Lewin (1963), Pendergast and Boag (1973), Moss (1977), Raveling (1979), Pulliainen and Tunkkari (1983), Thomas (1984) and Kehoe and Ankney (1985), suggest intestinal tract length increases and hypertrophy when an increase in dietary fiber occurs. The reported increase in intestinal length could mean merely an increase in food consumption, and the apparent hypertrophy due to inadequate removal of intestinal contents. Food intake is increased during the winter (Pendergast and Boag 1970), with a corresponding change to a high fiber diet in the birds studied (Davis 1961,

Pendergast and Boag 1973, Moss 1977, Raveling 1979, Kehoe and Ankney 1985). The increased food intake is likely due to both the winter conditions and the increased fiber in the diet; therefore the changes observed in intestinal length may not be due to dietary fiber per se.

Cecal measurements taken in the field could be influenced by time of day in the bird species where the ceca are more prominent. However, dark-eyed junco and Harris' sparrow ceca were not influenced by either collection time or diet, perhaps reflecting the diminished role of the ceca in the emberizids (Ziswiler and Farner 1972).

Gizzard weights changes reported by Spitzer (1972), Pendergast and Boag (1973), Miller (1975), Savory and Gentle (1976b), Raveling (1979), Hanssen (1979a), Paulus (1982), Drobney (1984), Herd and Dawson (1984), and Hupp and Brown (1984), cannot be explained by sampling time, grit, or inadequate content removal prior to weighing. Body weight (Moss 1983, Thomas 1984, Hobbaugh 1985), possibly influenced by sustained hypo- or hyperphagia (Raveling 1984, Hupp and Brown 1984) may explain some of the seasonal variation observed. Harris' sparrow gizzard changes may take a long time to develop, or may not occur at all.

Seasonal variation observed in the liver weights of

white-crowned sparrows (Oakeson 1953, 1956), Lagopus spp. (Thomas and Popko 1981, Pulliainen and Tunkkari 1984), sage grouse (Hupp and Brown 1984), snow geese (Ankney 1977), and wood ducks (Drobney 1979) might be partially explained by the time of day the birds were collected. Harris' sparrows did show liver size variations depending on the diet consumed however, and they were all sampled prior to feeding.

EXPERIMENT IV

FORMALIN EFFECTS ON DIGESTIVE ORGAN DRY WEIGHT

INTRODUCTION

Dry organ weights have not been taken before for any bird digestive morphology studies, although dry weight is considered more precise than wet measurements (Bowen 1983). If the more precise dry weights are to be taken, a means of storing the tissue prior to drying will be necessary, especially with field collection. Storage of the organ in formalin is expediant and inexpensive, but the effects of formalin preservation on dry weights needs to be addressed.

LITERATURE REVIEW

Adequate fixation and preservation occurs when the tissue is protected from self-digestion (autolysis) and bacterial attack (Sumner and Sumner 1969).

Formaldehyde, the active ingredient in formalin, achieves these goals by reacting with the amine groups on the amino acids (Steedman 1976) and possibly other reactive groups (French and Edsall 1945). Cytoplasmic proteins are thus turned into an insoluble macromolecular network (Jones 1976) that firms up the tissue and preserves cell structure (Sumner and Sumner 1969). Formaldehydes combination with active groups is slow (Barka and Anderson 1963) and also may be incompletely reversed (Pearse 1953).

Formalin penetrates the tissue and fixes it quickly (Krajian and Gradwohl 1952). Some shrinkage in the tissue due to muscular contractions may occur immediately (Drury and Wallington 1980) and for as long as two days later (Gabe 1976). Merriam (1959) found a 20 to 25% dry weight loss in formaldehyde preserved tissues, possibly due to the loss of soluble proteins and lipids. Contrarily, Jones (1976) found a 10 to 15% dry weight increase in formalin fixed tissues. Formalin induced dry weight changes likely vary depending on the pH, temperature, tissue, animal, or time of storage (Jones 1976, Steedman 1976).

METHODS

American tree sparrows (Spizella americana) were maintained in the laboratory at 10 C on a mash diet with a 10L:14D photoperiod (see general methods). Fourteen birds were separated into 7 pairs according to wing length and body weight. Pairs were sacrificed at the beginning of the photoperiod. One bird from each pair was randomly chosen to have its digestive organs immediately dried after evisceration, while the other bird had its organs fixed and preserved in formalin for 40 days prior to drying. All organs were dried at 65 C for 24 hours and weighed immediately thereafter.

RESULTS

Although data was collected in a paired manner, analysis was performed using pooled t-tests since the pairing criteria (wing length and body weight) did not provide for legitimate pairing ($r^2 = .12$ and $.15$, respectively).

Gizzard dry weight was significantly less when the gizzard was fixed (170.8 mg) instead of being dried immediately (193.9 mg)(Table 21). Gizzard wet weights were not significantly different between the two treatments (678.3 mg fixed and 671.1 mg dried immediately). Cecal dry weight was also significantly decreased when the ceca were put in formalin prior to drying (1.2 to 1.7 mg) instead of drying immediately after evisceration. Cecal wet weights were only slightly less in the organs to be fixed (6.0 to 6.2 mg).

Large intestine dry weights were also less when the organs were preserved first (6.4 to 10.5 mg), but this difference is compounded by lower wet weights in the large intestines to be fixed (27.3 to 35.9 mg) instead of dried immediately. Small intestine and liver dry weights were unaffected by formalin fixation.

Table 21. Dry weights (mg) for tree sparrow digestive organs either fixed in 10% buffered formalin and dried, or dried immediately upon evisceration.

	Not Fixed Prior To Drying	Fixed Prior To Drying	SE
Wing length (mm)	75.3	75.4	1.22
Body weight (g)	16.6	17.0	0.45
Small Intestine			
wet weight	726.7	654.6	51.43
dry weight	199.1	170.5	13.50
Large Intestine			
wet weight	35.9	27.3	5.45
dry weight	10.5 ^A	6.4	0.95
Ceca			
wet weight	6.2	6.0	0.77
dry weight	1.7 ^A	1.2	0.10
Gizzard			
wet weight	678.3	671.1	28.61
dry weight	193.9 ^A	170.8	6.78
Liver			
wet weight	606.9	656.0	34.41
dry weight	208.7	201.0	9.87

^ASignificant difference between weights, same organ.

DISCUSSION

The variable action of formalin on different organ dry weights corresponds with that reported by Steedman (1976). Unlike Jones (1976) no dry weight increases in digestive organs fixed in formalin were found, and when differences did occur, they followed the dry weight loss patterns indicated by Merriam (1958).

Why the small intestine and liver dry weights were not affected by formalin, while dry weights of the other digestive organs were, could not be explained by the surface area or size of the organ exposed. All organs, regardless of size were preserved for 40 days, a more than adequate time for complete fixation and preservation. The loss of lipids during formalin preservation (Merriam 1958) likely explains the differential dry weight loss observed. Since lipid levels change daily (Fisher and Bartlett 1957, Dolnick and Blyumental 1967) and seasonally (Odum and Perkinson 1951, Freeman 1969, Pulliainen and Tunkkari 1984) in different organs, formalin induced dry weight changes can be expected to occur differentially.

The use of formalin as a preservative for avian digestive tracts is not recommended if comparable dry weights to other studies is desired. Also, dry weights obtained throughout this study cannot be used to assume actual dry weights for comparison to other non-formalin

dry weights.

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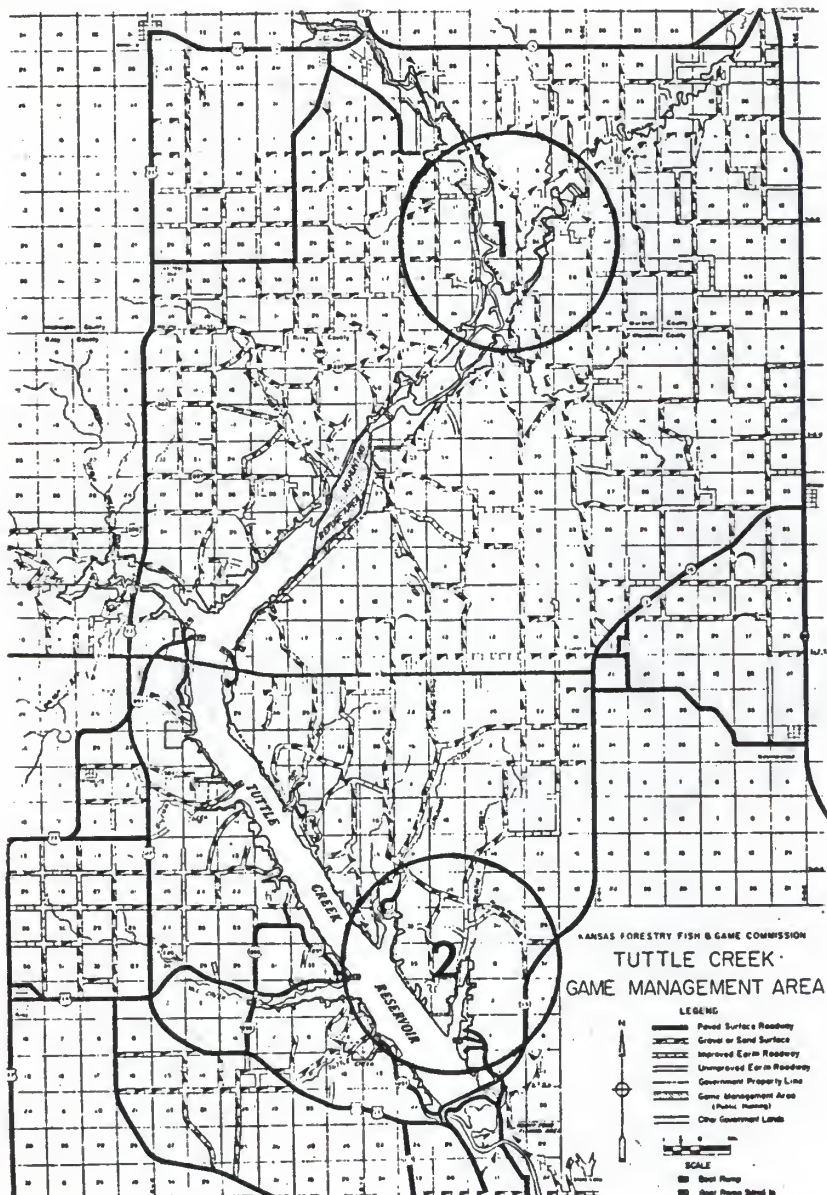
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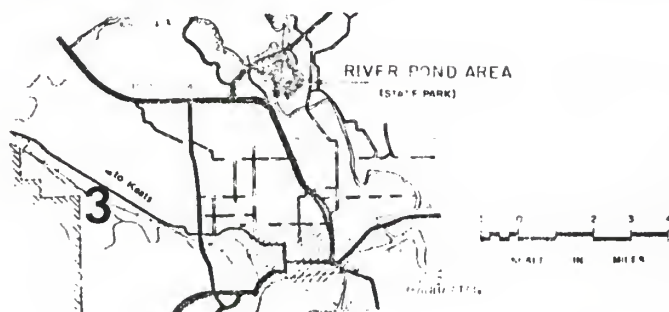
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Appendix A. Location of netting areas and primary species obtained.



Map 1. Tuttle Creek Area.

Map 2. Manhattan Area.



Site	Location	Map	Legal Description
1	Marshall County	1	NE1/4 Sect 31 T5S R8E Marsh. Co.
2	McIntire Cove	1	SE1/4 Sect 35 T8S R7E Pott. Co.
3	Blueville Nurs.	2	SW1/4 Sect 5 T10S R7E Riley Co.

Netting Success (10/86-2/87)

Location	Harris' Sp.	Junco	Tree Sp.	Cardinal
Marshall County	A	C	A	C
McIntire Cove	B	B	B	B
Blueville Nurs.	C	A	B	B
A = Excellent B = Good C = Poor				

Appendix B. Balanced mash (chick starter) formula that was prepared by the Kansas State University of Grain Science, and used as both a maintenance and experimental diet (from Schuman 1984).

Code No. P-17 Type of Feed Chick Grower

Formula date 11/8/77 Requested by Sanford Dept. Poultry Science

Date Mixed _____

INGREDIENTS

	AMOUNT/1000 lbs.	
	Individual	Cumulative
Bulk (pounds)		
Soybean oil meal (44%)	150	150
Ground yellow corn (8%)	250	400
Ground milo (9%)	240	640
Ground oats (13%)	100	740
Dehydrated alfalfa meal (19%)	50	790
Meat & bone meal (50%)	50	840
Fish meal (60%)	25	865
Wheat middlings (16%)	100	965
Premix A (pounds)		
Dicalcium phosphate	10	10
Limestone	10	20
Salt	5	25
Premix B (grams)		
Vitamin A (10,000 IU/g)	100	100
Vitamin D ₃ (15,000 IU/g)	20	120
Vitamin B ₁₂ (Proferm 20)	104	224
B-Complex (1233)	58	282
Amprol (25%)	227	509
Choline Chloride (50% mix)	400	909
Aurofac-10	208	1117
Trace minerals "CCC Z 5"	227	1344
Ground milo	3196	4540 (10 lbs.)

Services: Bulk _____ Paper bags X Burlap bags _____ Mix X

Pellet _____ Crumble X Grind _____ Other _____

Extrude _____ Compact _____ Conditioning temperature _____

Appendix C. Formalin used in the preservation and fixation of digestive organs.

**For In Vitro Diagnostic Use
For Tissue Fixation***

S0-F-99

**4 L
(1.1 gal.)**

**Specialty Filtered For Use In Fisher
Histomatic™ Tissue Processor**

Store at room temperature (20°-30°C)

Clear, colorless solution, otherwise discard.

Buffered with Sodium Acetate according to
A.C.P. Manual of Histology.

Reactive Ingredients 4% W/V
Sodium Acetate 2% W/V
Methanol 1.5% W/V

Stabilizer

LOT
860599-24
Expires
Feb. 1, 1988

Date Rec'd
/ /

**For laboratory and
household use only.
Not for drug, food, or
household use.**

CAS-50-00-0

Fair Lawn, New Jersey 07410
Made in U.S.A.

WARNING!

**CAUSES IRRITATION OF SKIN,
EYES, NOSE AND THROAT**

VAPOR HARMFUL

**CAUSES CANCER
IN LABORATORY ANIMALS**

Do not get in eyes. Avoid prolonged or repeated contact.
Avoid prolonged breathing of vapor. Use with ventilation.
In case of contact, immediately flush skin or eyes with
plenty of water for at least 15 minutes; for eyes, get
medical attention.

*Manual of Histological and Special Staining Techniques,
Second Edition, McGraw-Hill Book Co., New York, N.Y.,
1960, pages 1-2.

**10% Buffered
Formalin Acetate**

pH at 25°C 6.9-7.1

Certified

Fisher Scientific
An **ALLIED** Company

**Formaldehyde Solution
ORM-A UN2209**

**STORAGE
CODE**
B

**Fisher
ChemAlert™
Guide**

**NFPA
HAZARD
CODE**

HEALTH
Causes Cancer
In Laboratory
Animals

FLAMMABILITY

REACTIVITY

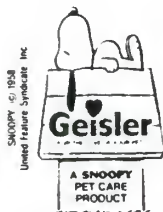
NO NFPA LISTING

**SAFETY
CODE**

EYE
GUARD
PROPER
GLOVES
SAFETY
CLOTHING

PURM H000

Appendix D. Formulation of a vitamin supplement that was given to Harris' sparrows during Experiment III, dietary and grit influences on digestive organ morphology.



drop-a-day

multi-vitamins for birds
WATER SOLUBLE

DROP-A-DAY is recommended as a multi-vitamin supplement to your bird's diet. It contains essential vitamins to aid birds that are confined to cages.

DIRECTIONS:

FOR NORMAL DOSAGE: Daily — one drop in drinking water and on food.

FOR AILING BIRDS: Daily — three drops in drinking water and three drops on food.

INGREDIENTS: Dextrose, Vitamin A Palmitate, D-activated Animal Sterol (source of Vitamin D₃), dl-alpha-Tocopheryl Acetate, Niacin Supplement, Calcium Pantothenate, Choline Bitartrate, Pyridoxine Hydrochloride, Thiamine Hydrochloride, Riboflavin-5-phosphate ester monosodium salt dihydrate, d-Biotin.

GUARANTEED ANALYSIS

Net 2.3 Fl. Oz.
19.7ml.

Vitamin A (Palmitate)	200,000 U.S.P.U.
Vitamin D ₃ (d-activated animal sterol)	40,000 U.S.P.U.
Vitamin E (dl-Alpha Tocopheryl Acetate)	30 I.U.
Thiamine	30 mg.
Riboflavin	20 mg.
d pantothenic acid	40 mg.
Niacin	150 mg.
Pyridoxine	8 mg.
Choline	24 mg.
d-Biotin	0.2 mg.
Sodium Benzoate — 0.1% added as preservative	

PET CARE BOOK

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Manufactured for:
ConAgra Pet Products Company
Omaha, NE 68105

PN73990P

Appendix E. Formulation of a mineral supplement that was given to Harris' sparrows during Experiment III, dietary and grit influences on digestive organ morphology.

LB
Lambert Kay

Amin

Liquid Mineral Supplement for Birds



Supplies the essential minerals needed for growth and reproduction. Soluble in drinking water; provides more complete mineral intake than powders & solids.

NET CONTENTS: 16 fl. oz. (473 ml)

	per ml.	per fl. oz.
calcium	13 mg	394 mg
sodium	6 mg	177 mg
zinc	250 mcg	7 mg
manganese	150 mcg	4 mg
copper	65 mcg	1.9 mg
iodine	1.5 mcg	44 mcg

INGREDIENTS: water, calcium borogluconate, sodium gluconate, calcium oxide, manganese gluconate, zinc gluconate, copper gluconate, potassium iodide, food grade coloring, potassium sorbate, methylparaben and propylparaben, as preservatives).

CAUTION: AMIN should not be stored or dispensed in galvanized containers.

LAMBERT KAY Div. of Carter-Wallace, Inc.
Cranbury, NJ 08512

PRODUCT #14202 LB-14202-05 Pat. Pend



0

Made & printed in U.S.A.

DIRECTIONS FOR DAILY USE

Mix as follows:

Dilute 1 part AMIN Liquid Mineral Supplement for Birds with 9 parts of water.

For breeding birds, dilute 1 part AMIN with 4 parts of water.

Fill drinking cup with solution.

AMIN may be mixed in drinking water with AVITRON Liquid Vitamin Supplement to supply both essential minerals and vitamins. When AMIN and AVITRON are mixed together, prepare a new solution at least every 3 days.

AMIN Liquid Mineral Supplement for Birds has been developed in collaboration with leading avian nutritionists to provide birds with the essential minerals necessary for reproduction, growth and development. Unlike powdered minerals, which are often discarded in the seed hulling process, AMIN is completely available in bird's drinking water for complete utilization and no waste.

Keep out of reach of children and pets. Use only as directed.

6A5704

EMBERIZID DIGESTIVE TRACT
LENGTH AND WEIGHT DYNAMICS

by

MICHAEL EDWARD BARNES

B.S., South Dakota State University, 1985

AN ABSTRACT OF A THESIS

submitted in partial fulfillment of the
requirements for the degree

MASTER OF SCIENCE

Division of Biology
KANSAS STATE UNIVERSITY
Manhattan, Kansas

1987

Dark-eyed juncos (Junco hyemalis), Harris' sparrows (Zonotrichia querula), and American tree sparrows (Spizella americana) were maintained in the laboratory at 10 C with a 10L:14D photoperiod. Juncos were used to determine sampling time effects on digestive organ morphology (gizzard, liver, both intestines and ceca), Harris' sparrows were used to test dietary effects, and tree sparrows were used to examine formalin effects on digestive organ dry weight.

Sex and age did not affect digestive organ morphology in dark-eyed juncos. In addition, no significant differences in any of the digestive organ morphological characteristics measured were found when the juncos were eviscerated up to 90 minutes after death. Both small intestine length and liver weight increased significantly throughout the photoperiod if free-feeding was allowed however. Slight diurnal decreases in small intestine length and large decreases in liver weight occurred in juncos deprived of food. Such diurnal variation in small intestine length could explain the observations reported in the literature. No other organ measurements were influenced by the time of day sampled.

Liver weights were the only digestive organ measurement that showed differences in Harris' sparrows fed either millet, sunflower or balanced mash. The lack

of dietary influence on intestinal length and weight was attributed to uniform sampling when the intestines were empty.

Gizzard and cecal dry weights decreased if the tissues were fixed in formalin prior to drying. Liver, small intestine, and large intestine dry weights were unaffected by formalin preservation.